

DRAFT

Working Group 6. Pediatric Liver Disease

Introduction & Background

Pediatric liver diseases include biliary atresia, metabolic disorders, intrahepatic cholestatic disorders, alpha-1-antitrypsin deficiency liver disease, nonalcoholic steatohepatitis (NASH), autoimmune hepatitis and sclerosing cholangitis, parenteral nutrition- and drug-induced liver injury, Wilson disease, cystic fibrosis and the various forms of viral hepatitis. While liver disease is uncommon in childhood, those liver diseases that occur tend to be severe and progressive. Many of these conditions are of unclear etiology and, for most, there are no effective medical therapies. Liver transplantation is an effective means of reversing acute liver failure or advanced end-stage liver disease in children and each year ~500 liver transplants are done in the U.S. in the pediatric age groups. While liver transplantation is often highly successful, the child then faces a life of managing the problems of immunosuppression and living with the transplanted organ.

The most common, severe liver disease of children is biliary atresia which affects ~1 in 10,000 newborns. Biliary atresia is characterized by a progressive inflammatory and fibrous obliteration of the large and intermediate intra- and extra-hepatic bile ducts. This leads to cholestasis, progressive liver fibrosis and cirrhosis. Without intervention, children die of end-stage liver disease within the first few years of life. The Kasai procedure (hepatoportoenterostomy), if done within the first few months of life, can partially restore bile flow and prevent liver failure. Nevertheless, more than two-thirds of children with biliary atresia will eventually require transplantation, and this diagnosis is the single most common reason for liver transplantation in children. The cause of biliary atresia is unknown; it presents soon after birth, but does not appear to be inherited. Hypotheses regarding the etiology of biliary atresia implicate perinatal infections, immune abnormalities and somatic mutations in laterality genes.

Other serious childhood liver diseases include neonatal hepatitis, three forms of progressive familial intrahepatic cholestasis (PFIC-1, -2 & -3), Alagille syndrome, alpha-1-antitrypsin deficiency, mitochondrial hepatopathies and several other inherited metabolic abnormalities. The genetic bases for some of these conditions are known, but many remain elusive, and there are no effective therapies for most, short of liver transplantation.

Of course, many of the liver diseases that affect adults can occur or have their onset in childhood. Prominent among these are hepatitis B and C, NASH, Wilson disease, autoimmune hepatitis and sclerosing cholangitis. Importantly, advances made in the prevention and treatment of these adult liver diseases have not always been applied to or assessed adequately in children. Differences in disease presentation, complications and progression make it difficult to extrapolate results of therapy in adults to children. In

particular, the implications of starting long-term, possibly life-long therapy for a chronic liver disease are quite different for children than for adults.

Recent Research Advances

Important advances have been made recently in the understanding of liver disease in children. Most strikingly, many of the genes of the inherited forms of liver disease in children have been identified, and the elucidation of the resulting biochemical and molecular defects promises to provide means of prevention and treatment of these diseases. Thus, the gene for Wilson disease has been shown to code for a copper-transporting ATPase that is required for normal copper excretion by the liver. The gene for Alagille syndrome has been shown to code for Jagged-1, which is a ligand in the Notch signaling pathway that is important in cell differentiation. The genes and the protein products of some forms of PFIC have been identified (FIC-1, BSEP, and MDR3), all of which appear to participate in bile secretion. The biochemical basis of hereditary tyrosinemia has been elucidated, and an inhibitor of tyrosine metabolism was found to reverse the metabolic abnormality and rescue infants with liver failure due to this disease.

Vaccines against hepatitis A and B have had major effects on the incidence of acute and chronic hepatitis in children. The use of universal hepatitis B virus vaccination has already decreased the frequency of chronic hepatitis B and liver cancer in China. Introduction of routine screening of blood for hepatitis B and C has eliminated post-transfusion and blood-product associated hepatitis, which were previously important causes of hepatitis in children. Nevertheless, pediatric cases of chronic hepatitis B and C are still seen, most as a result of maternal-infant transmission and/or emigration from areas of the world where these diseases are common. Therapy of hepatitis B and C has advanced substantially in the last 10 to 15 years. However, these therapies have been applied to only small numbers of children, and the optimal regimen, rates of response, predictive factors, and short- and long-term safety of these therapies have yet to be defined.

Due to the lack of effective medical therapies, liver transplantation has become the standard treatment for end-stage as well as fulminant liver disease in children. Advances in surgical techniques and immunosuppressive regimens have improved short- and long-term outcomes for children undergoing transplantation. Importantly, the use of reduced liver grafts, split livers and living donor livers has materially improved the prognosis of children with advanced liver disease, decreasing the waiting list mortality and providing better options for children facing the debility and burden of end-stage liver disease.

Research Goals

The major goals for research on pediatric liver diseases are to elucidate the causes of these diseases and develop practical means of their diagnosis, treatment and prevention.

Pathogenesis & Management of Biliary Atresia: The underlying causes of several important liver diseases in children remain elusive. Most importantly, the etiology of biliary atresia is unknown, and the search for the cause is a high priority (Matrix Cell C3). Approaches to elucidation of the cause of biliary atresia could include careful analysis of cohorts of patients with this disease and application of state-of-the-art means of assessing genes, gene regulation, proteins, lipids, environmental factors and infectious agents. Development of networks of interdisciplinary groups (clinicians, molecular biologists, geneticists, virologists) to study biliary atresia would help achieve this goal. The understanding of biliary atresia and many other pediatric liver diseases would be aided by a better understanding of normal organogenesis of the liver and biliary system during fetal life and a full characterization of the structural and functional changes that occur with development of the liver (Matrix Cell B1). Finally, clinical trials are important to ensure optimization of the medical and surgical management of biliary atresia with a focus on methods of early noninvasive diagnosis and ways to improve the success of the Kasai operation.

Molecular Pathogenesis & Treatment of Neonatal Cholestatic Syndromes: Despite identification of many of the genes that cause neonatal cholestatic liver disease, these advances have so far failed to result in practical means of prevention, treatment and cure of these diseases. Furthermore, the cause of many cases of neonatal cholestasis cannot be identified. Thus, many cases of PFIC have no mutations in *FIC-1*, *BSEP* or *MDR3*, and their underlying etiology remains unclear. Further investigation of the molecular pathogenesis of these diseases would help to define how the genetic defects lead to liver and biliary cell injury (Matrix Cells B2 and A1). Defining the molecular mechanisms by which these genes affect normal liver and biliary function would be aided by the development of animal models for these diseases (Matrix Cell B2). For genetic diseases, an approach using transgenic mice with the abnormal human gene would provide a means to better elucidate the mechanisms of cell injury. Animal models would also be important in providing insights into modifying genes and environmental influences in expression of disease as well as insights into therapeutic approaches. Importantly, animal models would provide the means to evaluate new therapies, which is exceedingly difficult for these relatively rare and severe diseases. Finally, animal models might allow for the development of practical means of molecular therapy of these inherited diseases, such as the use of small interfering RNA (siRNA), gene therapy, hepatocyte transplantation, or stem cell-based correction of metabolic abnormalities. New approaches to therapy may be possible, either using drugs or small molecules to help correct or bypass the loss or block of function (Matrix Cell C2) or by using molecular approaches, gene therapy or hepatocyte cell transfer (Matrix Cell C3). Clinical studies focusing on the natural history and management of patients with cholestatic syndromes are important. A major current handicap is the lack of noninvasive means for accurate diagnosis, staging and grading of disease. The evolving tools of genetics, genomics, gene

arrays, proteomics, and metabolomics could productively be applied to well-characterized patients to develop new hypotheses regarding pathogenesis, modifying factors and genes and to develop biomarkers for diagnosis, staging and grading of disease (Matrix Cells B2 and B3).

Characterization & Treatment of Pediatric Liver Diseases beyond the Neonatal Period:

The burden of liver disease in children in the U.S. is not well defined. Better systems could help to characterize the frequency and epidemiology of liver diseases in childhood and to help delineate their natural history (Matrix Cell A2). Clinical trials in children of antiviral agents for viral hepatitis that have been found to be effective in adults would help to define the optimal regimens of therapy, response rates, predictors of response, tolerability and safety. A major component of these studies should be assessing effects of therapy on growth, development and quality of life, as well as ancillary studies focusing on genetic modifiers of disease progression, noninvasive markers of fibrosis and early biomarkers for hepatocarcinogenesis. Nonalcoholic steatohepatitis (NASH) is an emerging liver disease, which is increasing markedly in frequency as a result of the recent increases in obesity in the United States. Originally described as a disease of middle-aged, overweight women with diabetes, NASH has now been shown to affect all age groups including young children. Clearly, clinical studies would enable researchers to characterize the clinical syndrome, natural history, course and complications of this disease in the pediatric age group (Matrix Cell A1). Ideally, these research studies would engage laboratory and basic researchers in attempts to define the etiology and pathogenesis of NASH and to provide insights into possible means of therapy. Clinical trials of new agents for NASH in adults should be matched with similar trials in children, with as little delay as possible.

Optimization of Liver Transplantation in Children: Liver transplantation is highly successful in children, but it still presents major challenges. Long-term outcome and complications are poorly characterized and require follow-up of large cohorts of carefully evaluated transplant recipients. Most long-term complications of transplantation are due to the immune suppression that is required to prevent rejection. A major challenge in liver transplantation is how to minimize immunosuppression without sacrificing graft function. Understanding the nature of immune tolerance, evaluating new tolerance-inducing regimens, delineating factors associated with tolerance, and developing biomarkers that accurately predict the necessary amount of immune suppression are all important issues that warrant prospective study (Matrix Cell B1). Emphasis should also be placed on means for avoiding the need for liver transplantation. For children with biliary atresia, liver transplantation may be avoided or at least delayed by early diagnosis, successful Kasai operation, and institution of effective anti-cholestatic and anti-fibrotic therapies thereafter. Research on biomarkers for early diagnosis and means to optimize the Kasai procedure are key to this goal (Matrix Cells B3 and C1). Acute liver failure of unknown cause is the second most common cause for liver transplantation in children. A major challenge for research is the identification of the etiology of cryptogenic acute liver failure in children (Matrix Cell A3) and developing both specific and nonspecific means of its prevention and amelioration (Matrix Cell C1).

Steps to Achieve Research Goals

Use of existing mechanisms as well as new ones will help to achieve the major research goals in pediatric liver disease. Thus, epidemiological systems that have been developed primarily for adult liver disease should be encouraged to include a component focusing on pediatric liver disease. Examples of such systems are the NHANES survey and the CDC surveillance systems for acute and chronic liver disease. Similarly, NIH-funded networks on liver disease in adults (such as those in NASH or autoimmune liver diseases) should include pediatric components, and studies of therapy in adults with chronic hepatitis B and C should be matched with studies in children. The newly funded cohort study on pediatric liver transplant recipients (SPLIT) could serve as an instrument for investigating long-term outcomes, complications and tolerance in children undergoing liver transplantation.

Advances in understanding of biliary atresia are likely to result from the newly formulated Biliary Atresia Research Consortium (BARC), which is a multicenter study focusing upon developing cohorts of well-characterized patients followed prospectively. An important role for BARC is to develop hypotheses regarding etiology and therapy of biliary atresia and help to test these hypotheses in patient material. BARC could also serve as a group to provide samples for development of biomarkers for the noninvasive diagnosis and staging of disease and to develop clinical trials focusing on optimizing management of biliary atresia. The advantages of a clinical research network in biliary atresia are many. Advances in research in other inherited cholestatic liver syndromes would be helped by expanding the BARC network to include other neonatal liver diseases such as Alagille syndrome, PFIC, alpha-1-antitrypsin deficiency, bile acid metabolism disorders and the mitochondrial hepatopathies. Understanding and management of these rare diseases would be greatly enhanced by focused attention and investigation by a multicenter network of investigators. Such an expanded network could provide serum samples, DNA materials and liver and biliary tissues for investigation and pursuit of etiology and pathogenesis; establish reliable definitions and diagnostic criteria for these diseases; and provide national centers of excellence for DNA-based diagnosis. This network might also establish resources and sources of support for patients and their families.

Benefits would flow from the placement of a continued emphasis on basic research on the liver as complementary to research on specific pediatric liver diseases. Of particular importance are studies of embryogenesis, organ development, angiogenesis, biliary cell differentiation, and hepatocyte function. Other important areas are stem cell research and gene therapy, including the development of better and safer methods for human gene and cell transfer. The development of animal models for biliary atresia and the neonatal cholestatic syndromes is of critical importance. Mechanisms would also be useful for supporting the development of new animal models, as well as the screening, identification and preclinical testing of small molecules for therapy and prevention.

Matrix of Research Goals in Pediatric Liver Disease

	Short-term Goals (0-3 years)	Intermediate-term Goals (4-6 years)	Long-term Goals (7-10 years)
High Risk	A3. Elucidate the major cause of idiopathic acute liver failure in children.	B3. Identify biomarkers for diagnosis, staging & grading of neonatal cholestatic syndromes.	C3. Define the etiology of biliary atresia. Develop gene, siRNA, cell transfer or stem cell therapy for pediatric metabolic disease.
Intermediate Risk	A2. Develop systems to better characterize the frequency, medical burden & epidemiology of pediatric liver disease.	B2. Delineate the molecular pathogenesis of at least 3 of the neonatal cholestatic syndromes. Develop better animal models for neonatal cholestatic syndromes.	C2. Based upon molecular pathogenesis (B2), identify small molecule therapies that might alleviate neonatal cholestatic syndromes.
Low Risk	A1. Characterize clinical syndrome, natural history, etiology, cofactors and complications of pediatric NASH. Develop definitions and diagnostic criteria for the major neonatal cholestatic syndromes.	B1. Define structural & functional development of liver/biliary system. Evaluate long-term outcomes, complications and tolerance-inducing regimens in children undergoing liver transplantation.	C1. Conduct clinical trials to optimize medical & surgical management of biliary atresia. Evaluate therapies for acute liver failure in children.